The Plant Cell, Vol. 31: 1393–1394, July 2019, www.plantcell.org © 2019 ASPB.

EDITORIAL: REFLECTIONS ON *THE PLANT CELL* CLASSICS

From Ethylene-Auxin Interactions to Auxin Biosynthesis and Signal Integration[OPEN]

The field of plant hormone biology, like many other research areas in plant sciences, has benefited tremendously from the adoption of Arabidopsis (*Arabidopsis thaliana*) as a model system. The development of a great genetic toolbox in this species led to the identification of many of the core components of the biosynthesis, signaling, and response pathways for all key plant hormones. With those components at hand, research emphasis progressively shifted toward understanding how the different hormones work in a coordinated fashion to orchestrate complex biological processes. This shift was further encouraged by the results of classical physiological and genetic experiments that clearly showed the importance of the interaction between multiple signals in a number of critical developmental processes. Among the most striking pieces of evidence for the existence of crosstalk between hormones were the repeated reports on well-characterized auxin mutants displaying reduced ethylene responses in roots. Despite these well-documented observations, the molecular mechanism behind these intriguing relations remained largely unknown until the mid 2000s.

In 2007, three articles were published back to back in *The Plant Cell* aiming to shed new light on the interplay of auxin and ethylene mediating root growth inhibition **(Růzicka et al., 2007; Stepanova et al., 2007; Swarup et al., 2007)**. Although approaching this question from different angles, all three articles reached the same general conclusion (i.e., an important part of ethylene effects on root growth is mediated by the stimulation of auxin biosynthesis and transport). While the contribution of specific auxin transporters was dissected in detail using existing mutants and reporter lines for the influx and efflux auxin carriers, at the time, only biochemical approaches with limited spatiotemporal resolution could be used to investigate the role of auxin biosynthesis in these interhormone relations. Thus, for example, the use of auxin and ethylene activity reporters allowed the authors of the three articles to establish specific roles for different parts of the root in this interaction process. The activity of an auxin-responsive reporter, *DR5*, in the root elongation zone was shown to be highly correlated with the ability of ethylene to inhibit root growth, whereas that was not the case in other regions of the root, such as the meristematic zone. Furthermore, the ethylene-triggered increase in the activity of the auxin reporter *DR5* in the elongation zone was also required for the activation of the ethylene reporter *EBS* in this part of the root. Altogether, the three articles provided a working spatiotemporal mechanistic model for the interaction between the two plant hormones. In this model, ethylene stimulates the production and transport of auxin toward the elongation zone of the root where this auxin, by a completely unknown mechanism, sensitizes the cells to the growth inhibitory effects of ethylene.

This mechanistic model has provided the basic conceptual framework not only to interpret physiological and genetic observations involving these two hormones but also for defining research questions that our laboratory and others have pursued. Thus, for example, the model advanced the idea that ethylene should stimulate local auxin production in the root, but it fell short of providing the specific mechanism. Follow-up studies described below led to the identification of a new family of auxin biosynthetic genes, the Trp aminotransferases *TAA1/TARs* (Stepanova et al., 2008), that, together with the previously identified flavin-containing monooxygenases *YUCCAs*, turned out to comprise the predominant auxin biosynthetic pathway in plants (Mashiguchi et al., 2011; Stepanova et al., 2011; Won et al., 2011). Similarly, this hormone interaction model also implied that certain threshold levels of auxin in the cells of the elongation zone were required for these cells' competence to respond to ethylene. Although possible mechanisms can be proposed at this time (see below), this is clearly an exciting area for future research in the field of hormone interactions.

One of the questions raised by the early studies was how ethylene stimulates auxin production. Characterization of two root-specific ethylene-insensitive mutants, *wei2* and *wei7*, with defects in the two subunits of anthranilate synthase, an enzyme involved in the production of an auxin precursor, amino acid ʟ-Trp, revealed a potential mechanistic link between ethylene response and Trp biosynthesis **(Stepanova et al., 2005)**. That report paved the way to another genetic approach that uncovered the link between ethylene and the biosynthesis of auxin (Stepanova et al., 2008). Conclusive genetic evidence for such a connection was established when the study of another root-specific ethylene-insensitive mutant, *wei8*, identified a previously uncharacterized aminotransferase, TAA1, that catalyzes the conversion of Trp to indole-3-pyruvic acid, the first committed step in the production of a major form of auxin, indole-3-acetic acid. Importantly, the *TAA1/TAR* family of aminotransferases was shown to be transcriptionally regulated by ethylene in the root meristem and in the elongation zone of the root (Stepanova et al., 2008). This finding not only pinpointed a mechanistic connection between ethylene signaling and auxin production but also was critical to the elucidation of the predominant indole-3-pyruvic-acid-dependent auxin biosynthetic pathway in plants (Mashiguchi et al., 2011; Stepanova et al., 2011; Won et al., 2011).

Another open question raised by the original ethylene-auxin interaction model is the nature of the molecular mechanisms by

---

[OPEN] Articles can be viewed without a subscription.

www.plantcell.org/cgi/doi/10.1105/tpc.19.00339

which auxin sensitizes a subset of root cells, specifically the cells in the root elongation zone, to ethylene. One can imagine a simple scenario in which auxin regulates the transcription of ethylene signaling components specifically in those cells. To our knowledge, no evidence for such regulation has been reported to date. Alternatively, auxin could control the activity of one or several ethylene signaling components at the posttranscriptional level. In fact, the levels of several ethylene signaling components have been shown to be regulated posttranscriptionally through the action of specific E3 ubiquitin ligase binding F-box proteins. Interestingly, the levels of two F-box proteins that target for degradation the transcriptional master regulators of the ethylene pathway, EIN3 and EIL1, are indeed translationally controlled (Li et al., 2015; Merchante et al., 2015) and modulated by auxin **(He et al., 2011)**. Whether or not auxin sensitizes the cells of the root elongation zone to ethylene by repressing the translation of *EBF* transcripts or by affecting EBF protein levels and/or activity by some other means, and how auxin regulates these processes, are interesting open questions that are still waiting to be answered. Finally, establishing a complete picture of these two hormones' relations in roots and other organs or tissues and developmental processes would require not only determining the complete set of molecular interactions involved but also deciphering the detailed map of spatial and temporal molecular events involved in the ethylene-auxin crosstalk. Some steps toward these ambitious goals have been recently taken by identifying the cells of the root epidermis as key players in the interaction between these two hormones (Brumos et al., 2018; Vaseva et al., 2018).

Anna N. Stepanova
Department of Plant and Microbial Biology
Program in Genetics
atstepan@ncsu.edu
ORCID ID: 0000-0003-1018-4758

Jose M. Alonso
North Carolina State University
Raleigh, North Carolina 27695-7614
jmalonso@ncsu.edu
ORCID ID: 0000-0001-7087-1571

ACKNOWLEDGMENTS

We thank Javier Brumos for the critical reading of the article.

REFERENCES

Brumos, J., Robles, L.M., Yun, J., Vu, T.C., Jackson, S., Alonso, J.M., and Stepanova, A.N. (2018). Local auxin biosynthesis is a key regulator of plant development. Dev. Cell **47:** 306–318.e5.

He, W., et al. (2011). A small-molecule screen identifies L-kynurenine as a competitive inhibitor of TAA1/TAR activity in ethylene-directed auxin biosynthesis and root growth in Arabidopsis. Plant Cell **23:** 3944–3960.

Li, W., Ma, M., Feng, Y., Li, H., Wang, Y., Ma, Y., Li, M., An, F., and Guo, H. (2015). EIN2-directed translational regulation of ethylene signaling in Arabidopsis. Cell **163:** 670–683.

Mashiguchi, K., et al. (2011). The main auxin biosynthesis pathway in Arabidopsis. Proc. Natl. Acad. Sci. USA **108:** 18512–18517.

Merchante, C., Brumos, J., Yun, J., Hu, Q., Spencer, K.R., Enríquez, P., Binder, B.M., Heber, S., Stepanova, A.N., and Alonso, J.M. (2015). Gene-specific translation regulation mediated by the hormone-signaling molecule EIN2. Cell **163:** 684–697.

***Růzicka, K., Ljung, K., Vanneste, S., Podhorská, R., Beeckman, T., Friml, J., and Benková, E.** (2007). Ethylene regulates root growth through effects on auxin biosynthesis and transport-dependent auxin distribution. Plant Cell **19:** 2197–2212.

***Stepanova, A.N., Hoyt, J.M., Hamilton, A.A., and Alonso, J.M.** (2005). A link between ethylene and auxin uncovered by the characterization of two root-specific ethylene-insensitive mutants in Arabidopsis. Plant Cell **17:** 2230–2242.

***Stepanova, A.N., Yun, J., Likhacheva, A.V., and Alonso, J.M.** (2007). Multilevel interactions between ethylene and auxin in Arabidopsis roots. Plant Cell **19:** 2169–2185.

Stepanova, A.N., Robertson-Hoyt, J., Yun, J., Benavente, L.M., Xie, D.Y., Dolezal, K., Schlereth, A., Jürgens, G., and Alonso, J.M. (2008). TAA1-mediated auxin biosynthesis is essential for hormone crosstalk and plant development. Cell **133:** 177–191.

Stepanova, A.N., Yun, J., Robles, L.M., Novak, O., He, W., Guo, H., Ljung, K., and Alonso, J.M. (2011). The Arabidopsis YUCCA1 flavin monooxygenase functions in the indole-3-pyruvic acid branch of auxin biosynthesis. Plant Cell **23:** 3961–3973.

***Swarup, R., Perry, P., Hagenbeek, D., Van Der Straeten, D., Beemster, G.T., Sandberg, G., Bhalerao, R., Ljung, K., and Bennett, M.J.** (2007). Ethylene upregulates auxin biosynthesis in Arabidopsis seedlings to enhance inhibition of root cell elongation. Plant Cell **19:** 2186–2196.

Vaseva, I.I., Qudeimat, E., Potuschak, T., Du, Y., Genschik, P., Vandenbussche, F., and Van Der Straeten, D. (2018). The plant hormone ethylene restricts *Arabidopsis* growth via the epidermis. Proc. Natl. Acad. Sci. USA **115:** E4130–E4139.

Won, C., Shen, X., Mashiguchi, K., Zheng, Z., Dai, X., Cheng, Y., Kasahara, H., Kamiya, Y., Chory, J., and Zhao, Y. (2011). Conversion of tryptophan to indole-3-acetic acid by TRYPTOPHAN AMINOTRANSFERASES OF ARABIDOPSIS and YUCCAs in Arabidopsis. Proc. Natl. Acad. Sci. USA **108:** 18518–18523.

*References highlighted for the 30th Anniversary of *The Plant Cell*.

From Ethylene-Auxin Interactions to Auxin Biosynthesis and Signal Integration

Anna N. Stepanova and Jose M. Alonso

Plant Cell 2019;31;1393-1394; originally published online May 8, 2019;
DOI 10.1105/tpc.19.00339